Pardaxin's action in shark

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OBJECTIVES

Structure and mode of action of the shark repellent pardaxin(Px)

ABSTRACT

A new column chromatography procedure, based on gel-permeation, ion exchange and chromatofocusing was employed to isolate the two main proteaceous, toxic, cytolytic and pore forming factors from the gland secretion of the Red Sea flatfish Pardachirus marmoratus. Pardaxin I(PXI) consisting 10% of the gland protein secretion, was shown to be 5-10 times more toxic, cytolytic and active on pore formation than pardaxin II (PXII) (8% of gland protein secretion). Gel-electrophoresis, amino acid analysis and N-terminal amino acid sequencing reveals a high degree of homogeneity and resemblance between the two toxins. They are rich in serine, glycine, alanine, leucine, and phyenylalanine and devoid of arginine and tryptophan. Their N-terminal was found to be NH_2-Gly-Phe-Phe. Their hydrophobicity is evident from the chrmoatographic behavior on a hydrophobic resin, presence of nine successive hydrophobic residues on the NH_2-terminal, overall percentage

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of toxins hydrophobicity and decreament of aqueous drops volume. The molecular weight of Pardaxin I in a monomer form is about 13 Kd as determined by gel filtration, gel electrophoresis and amino acid analyses. In aqueous solutions it tends to polymerize, mainly to a tetrameric form. Pardaxin I is composed of 120 amino acids and is free of carbohydrate and sialic acid residues. Mass spectrometry of the ethyl acetate extract of the gland secretion and the purified toxins reveals the presence of sterols in the secretion but their absence in the purified toxins.

Pardaxin I was iodinated without affecting its chemical and pore forming properties. It binds to liposomes, of different phospholipid compositions. In hyperpolarized unilamellar liposomes and multilamellar liposomes with an imposed chemical gradient, pardaxin I produced a fast, non,ion-specific (10^{-10}M) and slow, cation-specific pore (10^{-8}M) respectively (in preparation).

We have shown that in fish the gill tissue is the main organ for the action of PX. This action is being correlated with the channel-forming activity of PX. Our findings indicate that PX displays a channel or pore activity, e.g., it produces an increase in conductance in an artificial lipid membrane and in liposomes. The interaction of PX with fish gill tissue is being investigated presently.

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Moran, A., Korchak, Z., Moran, N. and Primor, N. Surfactant and channel-forming activities of the Moses Sole toxin. In, Toxins, drugs, and pollutants in marine animals. eds., L. Bolis, J. Zadunaisky and R. Gilles. Springer-Verlag Berlin, Heidelberg, New York, Tokyo. 1984 p. 13-15.

N. Primor. Pardaxin produces postjunctional muscle contraction in guinea-pig intestinal smooth muscle. Br. J. Pharmacol. 82, 43-49 (1984).

N. Primor, Pharyngeal cavity and the gills are the target organ for the repellent action of pardaxin in shark. Experientia. 41, 693-695 (1985).